



Genome Sequences of Two Novel Papillomaviruses Isolated from Healthy Skin of *Pudu puda* and *Cervus elaphus* Deer

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ABSTRACT We report the complete genome sequences of *Pudu puda papillomavirus 1* (PpudPV1) and *Cervus elaphus papillomavirus 2* (CelaPV2), isolated from healthy skin hair follicles of a Southern pudu and a red deer, respectively. PpudPV1 is basal to the DyokappaPVs, whereas CelaPV2 is basal to the XiPVs (Beta-XiPV crown group).

Papillomaviruses (PVs) are small nonencapsidated viruses with a circular double-stranded DNA genome of ca. 8 kbp in length. PVs infect epithelia in most amniotes, causing asymptomatic infections, proliferative benign lesions, and different cancers (1, 2).

Two adult deer living in captivity in zoos in Berlin, Germany, were sampled for the search of novel animal PVs. One of the specimens was a female Southern pudu (*Pudu puda*; Mammalia: Artiodactyla: Cervidae: Odocoileinae: *Pudu*). The Southern pudu is native to Argentina and Chile and is classified as “vulnerable” on the International Union for Conservation of Nature (IUCN) Red List of Threatened Species (<http://www.iucnredlist.org/>). The second specimen was a female red deer (*Cervus elaphus* Mammalia: Artiodactyla: Cervidae: Odocoileinae: *Cervus*). The red deer has a large global distribution extending from Europe and North Africa through central Asia, Siberia, the Far East and North America, and the IUCN Red List classifies this species as of “least concern.”

Hair follicles from the healthy skin of each animal were collected and tested for the presence of PV DNA using a battery of broad-spectrum PCR primers against the *E1* and *L1* genes (3). This partial information (ca. 450 bp) was used to design tail-to-tail primers to amplify the full-length genome using long-range PCR. The amplicons were Sanger sequenced in both strands through primer walking and cloned. Phylogenetic relationships for the *E1E2*, *L2L1*, and *E1E2L2L1* gene concatenates were inferred under a maximum likelihood framework (4).

Both *P. puda papillomavirus 1* (PpudPV1) and *C. elaphus papillomavirus 2* (CelaPV2) genomes show the classical PV genome arrangement: the upstream regulatory region (URR), the early genes *E6*, *E7*, *E1*, and *E2*, and the late genes *L2* and *L1*. The PpudPV1 *L1* gene shares similar levels of nucleotide identity with very divergent PVs: 66% with the giant panda *Ailuropoda melanoleuca papillomavirus 1* (AmPV1; Alpha-OmicronPV crown group: Omega PVs), 66% with the bottlenose dolphin *Tursiops truncatus papillomavirus 8* (TtPV8; Alpha-OmicronPV crown group, DyopiPVs), and 65% with the human PV 204 (HPV204; Kappa-LambdaPV crown group, MuPV). In accordance with the International Committee on Taxonomy of Viruses (ICTV) recommendations, this viral genome is thus of *incertae sedis*, clearly showing the need for strong revision of the

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current taxonomic criteria (5, 6). However, phylogeny confidently places PpudPV1 basal to the clade consisting of BPV18 and the DyokappaPVs. All members of this clade have been isolated from Caprinae (*Rupicapra rupicapra* [7] and *Ovis aries* [8]) and Bovinae species (*Bos taurus* [9, 10]), mostly from benign neoplasias. This PV clade does not belong to any of the four large PV crown groups (11).

The CelaPV2 *L1* gene shares between 67 and 69% nucleotide identity with different PVs in the XiPVs genus, all of them isolated from Bovidae and Cervidae species, which is in good agreement with the phylogenetic placement. CelaPV2 belongs thus in the Beta-XiPV crown group and is only distantly related to *C. elaphus papillomavirus 1* (Delta-ZetaPV crown group, EpsilonPVs), isolated from a red deer fibropapilloma (12, 13).

This study expands the known diversity of PVs infecting cervids and highlights the multiple evolutionary origins of PVs infecting cetartiodactyls.

Accession number(s). The complete genome sequences for PpudPV1 and for CelaPV2 are available in GenBank under the accession numbers [KT932713](#) and [KT932712](#), respectively.

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